Effects of Cover Plants on Soil Nutrient Status and on Growth of Hevea

I. Laboratory Studies on the Mineralisation of Nitrogen in Different Soil Mixtures

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The results of incubation studies on the mineralisation of nitrogen in soil, and in soil mixed with ammonium sulphate, calcium carbonate and plant material, are given. Only low levels of nitrification occurred in soil alone. Applications of ammonium sulphate and calcium carbonate, at levels equivalent to 4.3 cwt/acre and 133 lb/acre respectively, had little effect on mineralisation. Calcium carbonate applied at a rate equivalent to 4 tons/acre markedly stimulated nitrification. High levels of ammonium and nitrate nitrogen were found in mixtures of soil with Centrosema pubescens, Pueraria phaseoloides and Mikania scandens material, while none was found in mixtures of soil with hevea leaves and lalang (Imperata cylindrica).

CROP YIELDS ARE MORE UNIVERSALLY LIMITED by the level of available nitrogen in soil than by that of any other single nutrient. Available nitrogen consists of the ammonium and nitrate nitrogen contents of the soil and although there have been some reports that higher plants can assimilate nitrogen in the organic form (VIRTANEN & LINKOLA 1946, GHOSH & BURRIS 1950), it is this inorganic or mineral nitrogen which is the primary immediate source of the nutrient for all plants, other than the legumes and certain other nitrogen fixing plants. The mineral nitrogen constitutes only a small proportion of the total nitrogen in the soil; for instance, in a review of a number of experiments concerning annual agricultural crops, it has been reported that only about 3% of the total soil nitrogen was available in mineral form for uptake by the plants over a growing season of six months (BREMNER 1951). Because of this low level of available nitrogen, artificial nitrogenous fertilisers are used generally in the cultivation of annual crops with a high rate of demand for nitrogen in order to supplement the supply of available soil nitrogen. With tree crops, particularly those like hevea which grow under tropical conditions with practically a twelve month growing season, the rate of demand for nitrogen is slower and growth can be maintained at lower levels of available soil nitrogen than with most of the short season annual crops. In such instances the necessity for supplementary dressings of nitrogenous fertilisers decreases and the importance of the rate of release of available mineral nitrogen from the reserves of organic nitrogen in the soil increases.

The level of mineral soil nitrogen is governed by the well known nitrogen cycle. Plant and animal remains become incorporated in the soil and during their microbiological decomposition a certain proportion of their nitrogen content is released as ammonium nitrogen to the soil. This process, completed by the conversion of ammonium nitrogen into nitrate nitrogen, via the nitrite form, is termed the mineralisation of nitrogen. A proportion of the organic remains, including some nitrogenous compounds, is more resistant to decay, or becomes so in the process of decay, and contributes to the more stable organic matter and nitrogen reserves of the soil. Both ammonium and nitrate nitrogen can be taken up by plants and, in particular, this has recently been shown to be the case with hevea (BOLLE-JONES 1955). The

level of mineral nitrogen in the soil is governed by several factors; its rate of production is largely dependent on the degree of microbiological activity, depending as this does on the proportion of decomposable organic matter present to that of total nitrogen, and on the soil temperature, moisture, pH and other conditions. Once produced the ammonium nitrogen is subject to adsorption by the soil particles or conversion to the nitrate form. This nitrate can be quickly leached out of the soil by rainfall, or under certain conditions, a direct conversion of nitrate nitrogen to elemental nitrogen can result in a loss of gaseous nitrogen to the atmosphere.

Cultivation practices in hevea growing can greatly affect the natural mineralisation processes in the soil; clearing of the jungle in the first place exposes the soil to the sun with resultant increases in temperature and rate of mineralisation of the organic matter reserves of the soil. The use of cover plants helps to restore the equilibrium by maintaining cool and moist surface conditions, and the return of dead plant material to the soil by these plants provides a source of readily decomposable organic matter which can release nitrogen for uptake by the rubber tree. Ammonium sulphate fertiliser is generally used in hevea plantations to supplement the reserves of soil nitrogen, but it is important to understand the main characteristics of the natural nitrogen cycle in the soil and to determine in what way cultivation practices can be modified to ensure the maintenance of satisfactory levels of available soil nitrogen.

This is the first of a series of papers on this subject and gives the results of laboratory investigations which have been carried out while a series of field experiments is being laid down.

EXPERIMENTAL

The mineralisation of nitrogen, both in soil as received from the field and in soil to which plant material or ammonium sulphate has been added, and the effect of additions of lime to the soil on this mineralisation have been studied.

Samples representing three main types of soil on which hevea is grown in Malaya were used for the work. Brief particulars of these soils, together with the results of their chemical analysis are given in Table 1.

Methods

The standard bottle incubation method was used to study the mineralisation process. Three hundred grammes of soil were weighed in a basin and sufficient water was added to bring the soil moisture content to one third of the saturation capacity. The soil was mixed well and transferred into a 1 lb wide necked bottle. The bottle was corked loosely to reduce loss of moisture by evaporation and to protect the sample from contamination while allowing some movement of air, and was then incubated at a constant temperature of 30°C (±1°C). Once a week the soil was thoroughly mixed and samples were taken out for analysis. At each sampling the slight loss of moisture caused by evaporation from the soil while in the incubator was made up by addition of water. Two portions of incubated material were taken at each sampling, the amount depending on the expected content of ammonium and nitrate nitrogen, and these samples were extracted for ammonium and nitrate nitrogen separately.

For the estimation of ammonium nitrogen, the sample was shaken with an appropriate amount, depending on the ammonium nitrogen content, of N K_2SO_4 solution made 2N with respect to H_2SO_4, and filtered. The ammonium content of the filtrate was determined, in duplicate, by Conway's micro-diffusion method (Bremner & Shaw 1955).

Nitrate nitrogen was extracted with dilute calcium sulphate solution. If the extract was clear and colourless the nitrate was determined by the phenoldisulphonic acid

colorimetric method using a Spekker absorptiometer. In coloured extracts, generally obtained from samples containing high levels of decomposing organic matter, the nitrate was determined by Conway's micro-diffusion method using Devarda's alloy and magnesium oxide. Both methods had been shown in preliminary work to give identical results and their combined use lent some flexibility to laboratory arrangements.

Total nitrogen and organic carbon contents in the soil were determined by the normal micro-kjeldahl and Walkley and Black's wet digestion methods respectively. The organic nitrogen content of the soil was calculated by subtracting the mineral nitrogen content from the total nitrogen content; the resultant values have been used in all considerations of the C:N ratios of soils.

RESULTS

Mineralisation of Nitrogen in Soil as Received from the Field, and the Effect of Added Ammonium Sulphate and Calcium Carbonate on this Mineralisation

Samples of the three soils described above were incubated for seventy days. Analysis of weekly samples gave the results shown in *Figure 1*. It can be seen that, with each soil, higher levels of ammonium nitrogen than of nitrate nitrogen were found with each sampling. One marked feature, occurring with each soil, was the sharp increase in ammonium content during the first fortnight of incubation; with soil S_3 the level of ammonium content continued to rise steadily after the first fortnight whilst only a slight increase could be recorded with soil S_1, and with soil S_2 an actual decrease in ammonium content set in after about five to six weeks. This variation appears to be connected to the relative rates of nitrification occurring in the three soils; S_3 exhibited no nitrification at all over the experimental period, S_1 showed a slow rate of nitrification whilst a steady rate of nitrification was found with S_2 after the first week of incubation.

In another series of incubations ammonium sulphate was mixed with the soil at the rate of 5 mg of nitrogen per 100 gm soil, equivalent to a field application of 4.3 cwt/acre of ammonium sulphate (assuming 2 million lb soil per acre/six inches). The results given in *Figure 1* show that the basic mineralisation processes of the three soils were not altered; levels of ammonium nitrogen were higher by approximately 5 mg over the whole incubation period, and it was only with soil S_2 that nitrification appeared to be affected, a slight depression in the level of nitrate nitrogen produced being recorded, although the rate of nitrification was unaltered.

Application of calcium carbonate to the soil at a rate equivalent to 133 lb/acre caused no appreciable effect on the mineralisation process. When mixed in with the soil at a rate equivalent to 4 tons/acre, however, calcium carbonate caused marked effects on the mineralisation noted above. These effects are shown graphically in *Figure 2*. Comparison with *Figure 1* shows that with soils S_1 and S_2, no effect on the initial ammonification was caused by the calcium carbonate, but nitrification in these soils was so stimulated after an incubation period of two to three weeks that higher levels of nitrate nitrogen than of ammonium nitrogen were found after a further three weeks' incubation. This stimulation of the rate of nitrification ceased after another two to three weeks, when levels of ammonium and nitrate nitrogen in the soil levelled out.

Different effects were found with soil S_3; the initial increase in ammonium content found with the soil incubated alone was stimulated by the calcium carbonate, and vigorous nitrification was developed in the soil only after an incubation period of nine weeks. This soil showed no nitrification at all when incubated in the absence of calcium carbonate and it appears that a distinct difference exists between its nitrifying capacity and that of the other two soils.

The Effect of Added Plant Material on the Mineralisation of Nitrogen

The aerial portions (leaves, twigs and stems) of different cover plants which occur in hevea plantations were dried, ground and analysed (Table 2). Samples of these plant materials were mixed with samples of all three soils in the ratio of one to ten. The mixed soil and plant material was incubated as usual and samples were withdrawn for analysis at weekly intervals over a period of seventeen weeks. The results of analysis for ammonium and nitrate nitrogen in tests using material from the leguminous creepers *Centrosema pubescens* and *Pueraria phaseoloides*, and from the non-leguminous creeper *Mikania scandens* are given in *Figure 3*.

It can be seen that major differences occur between the mineralisation of the *Centrosema pubescens*/soil mixture and that of the *Mikania scandens*/soil mixture. In the former, a high rate of mineralisation produced initially very high levels of ammonium nitrogen reaching a peak equivalent to 100 mg nitrogen per 100 gm mixture, which commenced to fall after a time as vigorous nitrification developed (particularly with soils S_1 and S_2). With *Mikania scandens* mixture a much slower rate of mineralisation produced peak levels of ammonium nitrogen equivalent to only 30 mg nitrogen per 100 gm mixture. The subsequent nitrification in the *Mikania scandens* mixture was also at a lower level, except for a rather anomalous effect with soil S_3 in which slightly higher levels of nitrate nitrogen were produced in the *Mikania scandens* mixture than in the *Centrosema pubescens* mixture.

Ammonification in the *Pueraria phaseoloides*/soil mixtures was lower with each soil type than in the corresponding *Centrosema pubescens*/soil mixtures, particularly with soil S_2 and S_3. With soil S_2 and S_3 higher nitrification was observed in the *Pueraria phaseoloides*/soil mixtures than in the *Centrosema pubescens*/soil mixtures, while with soil S_1 there was little difference between the two mixtures.

Incubations of mixtures of lalang (*Imperata cylindrica*) and senescent hevea leaves with soils were also carried out, but in these instances no mineral nitrogen at all was produced during incubation.

The carbon and nitrogen contents and the C:N ratios of the untreated soils before incubation are given in Table 1, while those of the samples after incubation with or without applied ammonium sulphate and/or calcium carbonate are shown in Table 3. Though the three soils differ considerably as to their organic carbon and nitrogen contents, the C:N ratios both before and after incubation varied only between 9.00 and 11.9. This similarity, together with the low rate of decomposition, leads one to suppose that the soils contained only low reserves of decomposable organic matter.

The pH values of the soils alone, of soils treated with ammonium sulphate equivalent to 4.3 cwt/acre and/or calcium carbonate equivalent to 4 tons/acre, and of the soil/plant material mixtures were determined both before and after incubation and are shown in Table 3. The C:N ratios of the plant materials alone are given in Table 2 and those of plant/soil mixtures are included in Table 3. The latter also summarises the total nitrogen mineralised, in actual amount and as a percentage of the original nitrogen, calculated from the total amount of ammonium and nitrate nitrogen produced during incubation of these mixed samples. It can be seen that the mineral nitrogen produced during incubation of *Mikania scandens* mixtures is much lower, both in amount and in percentage of the original organic nitrogen, than that produced in *Pueraria phaseoloides* or *Centrosema pubescens* mixtures.

DISCUSSION

Three main points that arise from the above work call for discussion. They are the variation in nitrifying capacities of the three soil types used, and the effects of added calcium carbonate and plant materials respectively on the mineralisation process.

The ammonification and nitrification processes in the three untreated soils during incubation are typical of soils of high acidity and moderate organic matter content (PAUL & SHARIFF 1954, STOBE 1952, DUCHAUFOUR 1954). A comparatively high level of ammonification was followed by the development of only a low level of nitrification. It appears possible that even within the small range of pH values covered by these three soils, the degree of nitrification was related to the soil reaction — the soil with the lowest pH value (S_3) exhibiting the lowest rate of nitrification — and that the slow rates of nitrification in these soils, as compared to that in nearly neutral soils (ACHARYA & JAIN 1954), was the direct result of their low pH values. A greater number of soils with differing acidities would need to be studied before a definite conclusion on the above could be made, however. Doubtless the environmental conditions of the soils in the field will have exerted some effect; for instance soil S_3 was taken from shaded conditions under mature hevea whilst soils S_1 and S_2 came from replanting sites that had been exposed to the sun for some time, with possible consequences on their microbial population.

Though the organic nitrogen contents of the three untreated soils differed considerably, the amounts of nitrogen mineralised during incubation were all low and similar and were not related to the respective C:N ratios determined both before and after incubation. This is not surprising since variation in the histories of the soils will have produced differences in the soil organic matter such that the values of the C:N ratio at which only low values of mineralisation of nitrogen are found will vary from soil to soil (ANDERSON & BYERS 1934, KOHNLEIN 1955, ALBAREDA 1955).

The addition of ammonium sulphate to the soils only slightly affected the soil reaction (Table 3) and no significant effects on the mineralisation process were noted. The addition of calcium carbonate to the soil at a rate equivalent to 133 lb/acre, intended to affect only the nutrition of possible nitrifiers (STEVENSON & CHASE 1953) without appreciable change in pH value of the medium, caused no significant increase in the rate of nitrification. However, the addition of calcium carbonate at a level equivalent to 4 tons/acre caused major increases in both pH values and in nitrification, and it would appear likely that this is an instance where a decrease in acidity has stimulated development of the nitrifying population (MULDER 1950, CORNFIELD 1952, 1953, MAZILKIN 1954). The stimulation of nitrification took longer to develop with soil S_3 than with the other two soils. This may be related to the absence of nitrification found in the incubation of this soil when untreated. A microbiological examination of the soils might show that soil S_3 possessed an initially much smaller population of nitrifiers than did the other two soils.

In further tests, additions of calcium carbonate equivalent to a rate of 9 tons/acre caused no additional increase in nitrification over that produced by the dressing of 4 tons/acre; this agrees with other work showing that increases in pH values of over 6.5 by additions of calcium carbonate did not result in further increases in nitrification (CORNFIELD 1952).

In connection with this effect of pH on nitrification, it has been shown that fungi are predominant in the microbiological population of Malayan acid soils (CORBETT 1934); fungi are known to be effective ammonifiers (WAKSMAN 1952) and this combination of facts may explain why, in the untreated soils, ammonification was at a higher level than nitrification.

Admixtures of *Pueraria phaseoloides*, *Centrosema pubescens* and *Mikania scandens* to all three soils resulted in high rates of ammonification and subsequent nitrification, with *Pueraria phaseoloides* having the greatest effect and *Mikania scandens* the least. The variation in the amounts of nitrogen mineralised must have been dependent to some extent on the nature and ease of decomposition of the different plant materials and not primarily on their nitrogen contents nor on their C:N ratios, for the differences

between the three plant materials with respect to these factors seem hardly large enough to explain adequately the comparatively low level of mineralisation shown by the *Mikania scandens* (Table 3). Neither senescent rubber leaves nor lalang with C:N ratios respectively of 27.9 and 61.2 showed any mineralisation at all when mixed with soil, in the time covered by the experiment; this fact is in line with the theory that only material with a C:N ratio of 20:1 or lower can, in general, directly provide mineral nitrogen (HARMSEN & VAN SCHREVEN 1955).

The addition of some plant materials caused even greater increases in nitrification than that caused by the addition of calcium carbonate. Some intermediate determinations showed that, during the initial ammonification, the pH values of incubated samples increased until nitrification commenced, but fell again with the production of nitrate. The ammonification and nitrification effects in the soil/plant material mixtures are reflected in the change of pH values shown in Table 3. It is possible that nitrification was initiated in the soil/plant material mixtures by the decrease in acidity consequent on ammonification; the presence of ammonium nitrogen in itself was not sufficient to stimulate nitrification, for the addition of ammonium sulphate to soil had no effect on nitrification nor on pH value. This may explain why in some acid soils ammonium nitrogen from ammonium hydroxide or anhydrous ammonia is more rapidly nitrified than that from ammonium sulphate (ENO & BLUE 1954, BIRECKA et alii 1954, ANDERSON & PURVIS 1955).

It can be tentatively concluded from this preliminary work that, in bare soil containing only low levels of easily decomposable organic matter, the mineralisation of soil nitrogen is likely to consist largely of ammonification, only a low level of nitrification being attained. The rate of nitrification can be stimulated by raising the pH values of the soil by addition of calcium carbonate while the addition of large quantities of decomposable organic matter will stimulate both ammonification and nitrification. In young and mature plantations where such organic matter is returned to the soil as litter from cover plants comparatively high rates of ammonification and nitrification may exist. In mature plantations where hevea leaves form the principal leaf litter, it seems unlikely that rapid mineralisation of the leaf nitrogen content can occur.

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TABLE 1. CHEMICAL ANALYSIS OF SOIL SAMPLES

Results are calculated on oven dry basis

Soil	Site	pH	Ammonium nitrogen p.p.m.	Nitrate nitrogen p.p.m.	Organic nitrogen %	Organic carbon %	Organic carbon / nitrogen	Exchangeable cations (m.eq. per 100 gm soil) Ca	Mg	K
S_1 — a coastal alluvial clay-loam (Selangor series, Owen 1951)	Old hevea plantation cleared for replanting	5.09	30	35	0.295	2.73	9.25	3.08	3.17	0.96
S_2 — a clay loam containing a high proportion of coarse quartz granules (Rengam series soil)	Young hevea plantation; undulating ground lightly covered by weeds and grasses	5.39	39	5	0.166	1.92	11.57	1.19	0.06	0.32
S_3 — a sandy loam derived from sandstone. (Kedah series)	Mature hevea plantation; undulating ground supporting indigenous mixed cover plants	4.59	trace	trace	0.139	1.25	8.99	0.08	0.07	0.10

TABLE 2. CHEMICAL ANALYSIS OF PLANT MATERIALS

Results are calculated as percentages on oven dry basis

Plant material	Carbon	Nitrogen	Carbon/Nitrogen
Pueraria phaseoloides	41.70	2.91	14.3
Mikania scandens	38.76	2.44	15.9
Centrosema pubescens	40.32	3.53	11.4
Imperata cylindrica (lalang)	45.28	0.74	61.2
Senescent hevea leaves	42.40	1.52	27.9

TABLE 3. CHANGES IN ORGANIC CARBON, NITROGEN, CARBON/NITROGEN RATIOS AND pH VALUES DURING INCUBATION

Carbon and nitrogen results are expressed as percentages on oven-dry basis.

Value I = before incubation II = after incubation III = I – II		Soil alone	Soil + ammonium sulphate	Soil + calcium carbonate	Soil + ammonium sulphate + calcium carbonate	Soil + *P. phaseoloides*	Soil + *M. scandens*	Soil + *C. pubescens*
Soil S_1								
Organic carbon	I	2.73	2.73	2.73	2.73	6.31	5.93	6.11
	II	2.74	2.70	2.79	2.71	4.23	4.62	4.43
	III	+ 0.01	– 0.03	+ 0.06	– 0.02	– 2.08	– 1.31	– 1.68
Organic nitrogen	I	0.295	0.295	0.295	0.295	0.531	0.475	0.587
	II	0.281	0.285	0.270	0.270	0.476	0.433	0.457
	III	– 0.014	– 0.010	– 0.025	– 0.025	– 0.055	– 0.042	– 0.130
Carbon/ nitrogen ratio	I	9.25	9.25	9.25	9.25	11.88	12.48	10.40
	II	9.75	9.47	10.33	10.04	8.89	10.67	9.69
	III	+ 0.50	+ 0.22	+ 1.08	+ 0.79	– 2.99	– 1.81	– 0.71
Amount and percentage of N mineralised		0.012 (4.1%)	0.012 (4.1%)	0.017 (5.8%)	0.016 (5.4%)	0.075 (14.1%)	0.025 (5.3%)	0.100 (17.0%)
pH	I	5.09	4.75	5.83	5.78	5.32	4.38	5.06
	II	4.40	4.34	4.74	4.91	4.60	5.62	4.51
	III	– 0.69	– 0.41	– 1.09	– 0.87	– 0.72	+ 1.24	– 0.55

Soil S_2								
Organic carbon	I	1.92	1.92	1.92	1.92	5.43	5.07	5.26
	II	1.85	1.78	1.67	1.76	3.44	3.37	3.67
	III	− 0.07	− 0.14	− 0.25	− 0.16	− 1.99	− 1.70	− 1.59
Organic nitrogen	I	0.166	0.166	0.166	0.166	0.401	0.349	0.449
	II	0.156	0.152	0.138	0.151	0.379	0.336	0.355
	III	− 0.010	− 0.014	− 0.028	− 0.015	− 0.022	− 0.013	− 0.094
Carbon/ nitrogen ratio	I	11.57	11.57	11.57	11.57	13.54	14.53	11.71
	II	11.86	11.71	12.10	11.67	9.08	10.03	10.34
	III	+ 0.29	+ 0.14	+ 0.53	+ 0.10	− 4.46	− 4.50	− 1.37
Amount and percentage of nitrogen mineralised		0.013 (7.8%	0.012 (7.2%)	0.015 (9.0%)	0.012 (7.2%)	0.060 (15.0%	0.013 (3.7%	0.040 (8.9%)
pH	I	5.39	5.31	7.16	7.28	5.27	5.35	5.44
	II	4.70	4.52	6.65	6.10	4.65	5.50	5.32
	III	− 0.69	− 0.79	− 0.51	− 1.18	− 0.62	+ 0.15	− 0.12
Soil S_3								
Organic carbon	I	1.25	1.25	1.25	1.25	4.77	4.42	4.61
	II	1.37	1.31	1.28	1.37	3.06	2.84	3.04
	III	+ 0.12	+ 0.06	+ 0.03	+ 0.12	− 1.71	− 1.58	− 1.57
Organic nitrogen	I	0.139	0.139	0.139	0.139	0.373	0.322	0.425
	II	0.126	0.123	0.121	0.122	0.318	0.294	0.238
	III	− 0.013	− 0.016	− 0.018	− 0.017	− 0.055	− 0.028	− 0.187
Carbon/ nitrogen ratio	I	8.99	8.99	8.99	8.99	12.78	13.73	10.85
	II	10.87	10.65	10.58	11.23	9.62	9.66	12.77
	III	+ 1.88	+ 1.66	+ 1.59	+ 2.24	− 3.16	− 4.07	+ 1.92
Amount and percentage of nitrogen mineralised		0.014 (10.17%)	0.013 (9.4%)	0.015 (10.4%)	0.014 (10.1%)	0.070 (18.8%)	0.008 (2.5%)	0.055 (12.9%)
pH	I	4.59	4.70	6.91	7.11	4.77	5.43	5.15
	II	5.72	5.05	5.40	5.15	5.01	5.75	5.90
	III	+ 1.13	+ 0.35	− 1.51	− 1.96	+ 0.24	+ 0.32	+ 0.75

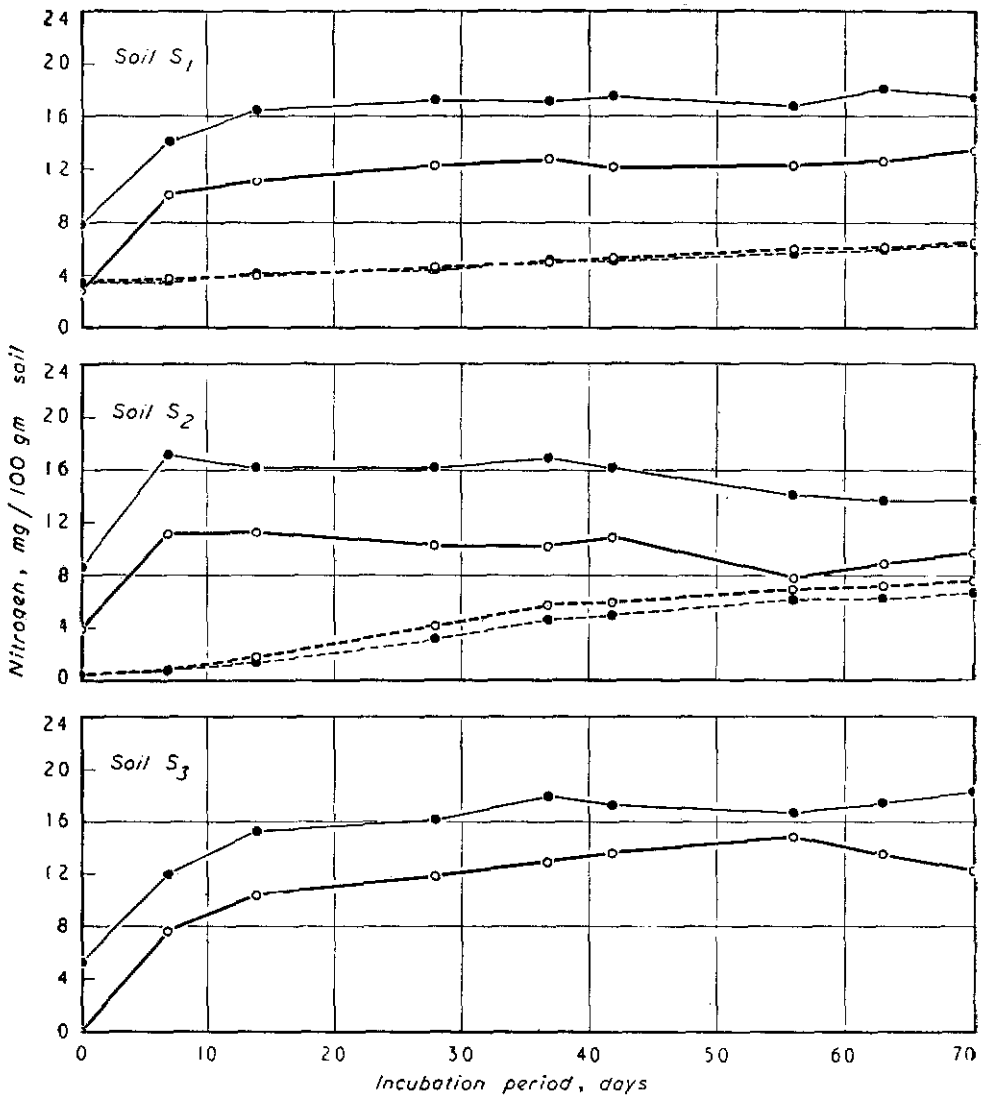


Figure 1. Mineralisation of nitrogen in soil alone and in soil mixed with ammonium sulphate.

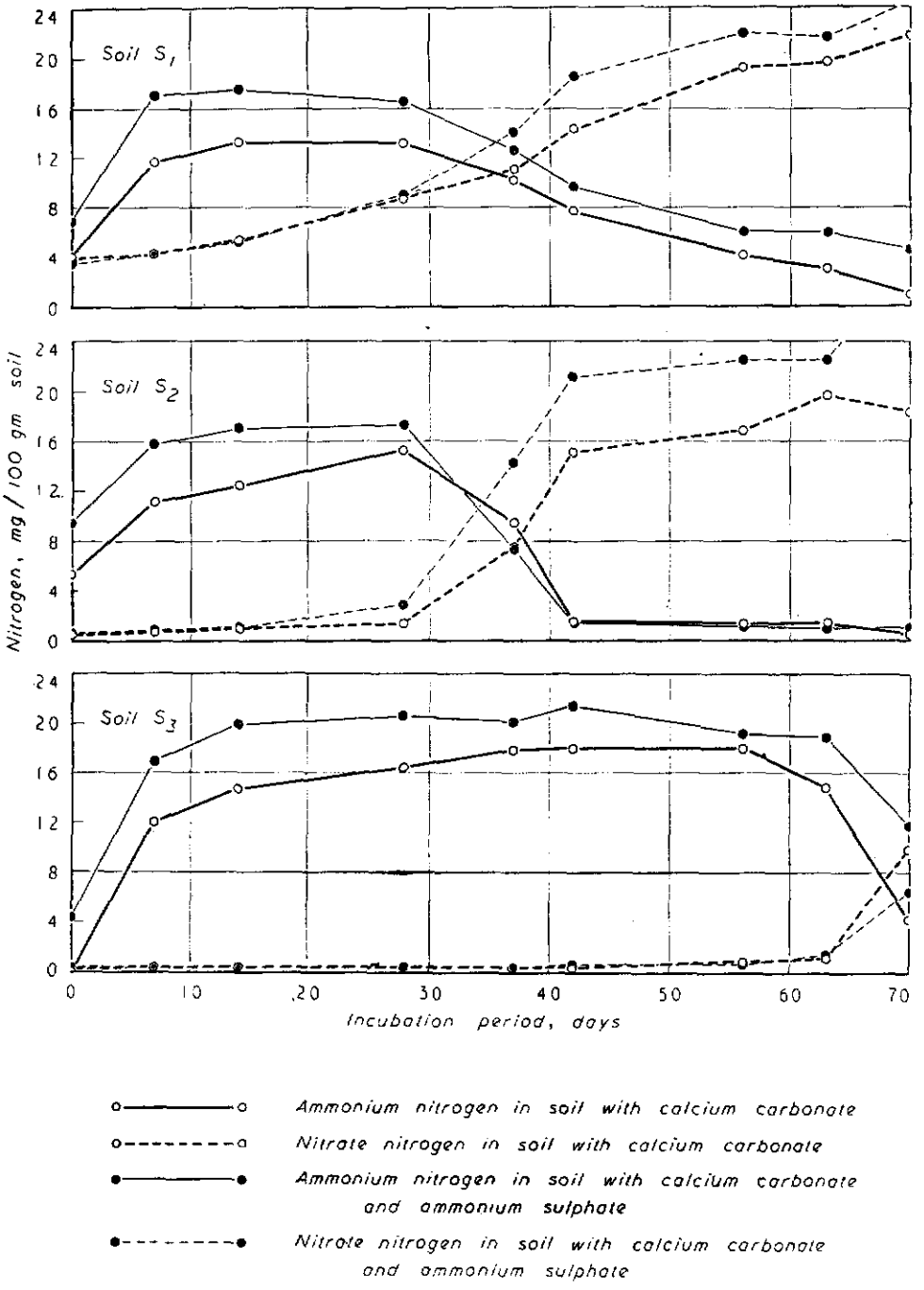
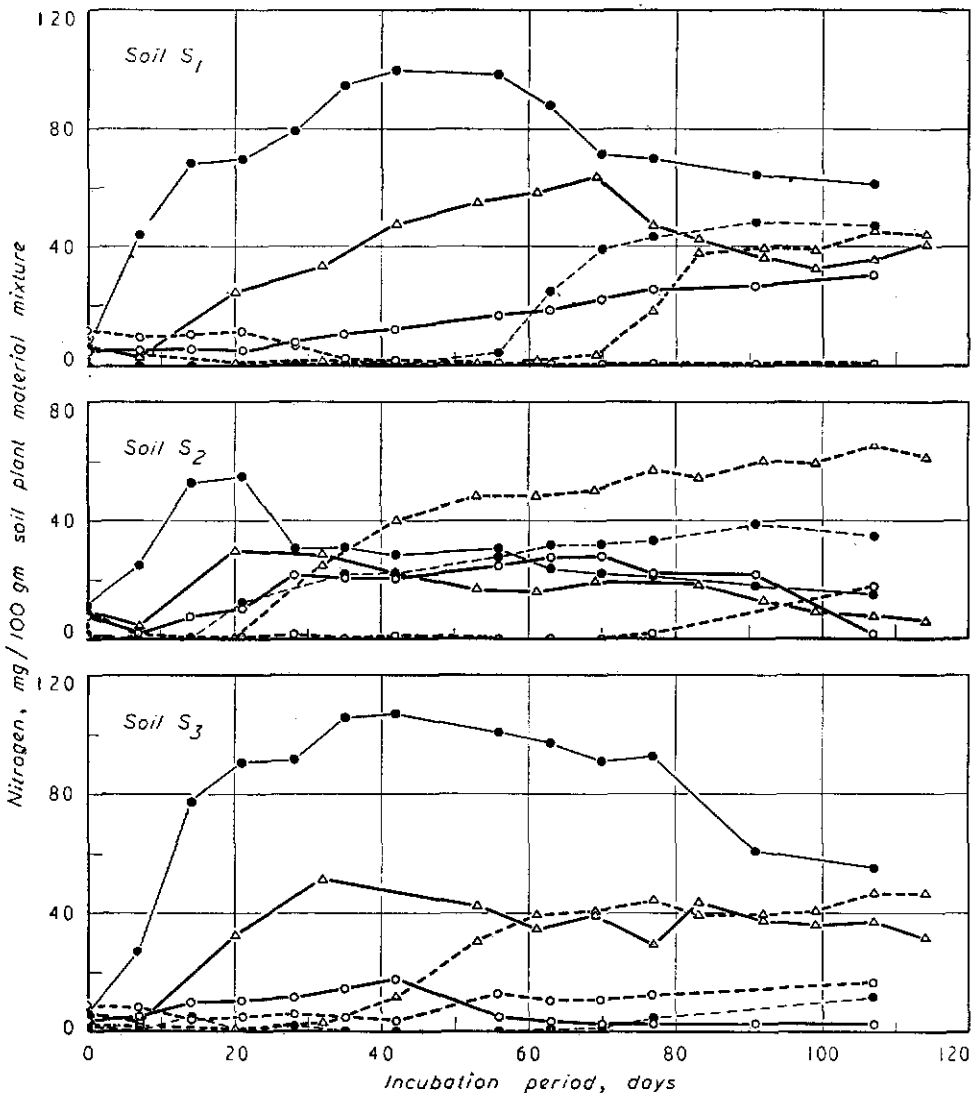


Figure 2. Mineralisation of nitrogen in soil mixed with calcium carbonate, and with calcium carbonate and ammonium sulphate.



Ammonium nitrogen in soil with Mikania scandens

Nitrate nitrogen in soil with Mikania scandens

Ammonium nitrogen in soil with Centrosema pubescens

Nitrate nitrogen in soil with Centrosema pubescens

Ammonium nitrogen in soil with Pueraria phaseoloides

Nitrate nitrogen in soil with Pueraria phaseoloides

Figure 3. Mineralisation of nitrogen in soil mixed with Mikania scandens, Centrosema pubescens and Pueraria phaseoloides material.